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STATISTICAL STUDY, CONTROL AND PREVENTION OF DIFFERENT NEWCASTLE DISEASE DIAGNOSED IN THE WESTERN OF ALGERIA

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Abstract. This study investigates avian viral infections, with a particular focus on Newcastle disease, across several farms in Algeria. A statistical model is proposed to address and characterize this phenomenon. A multiple-choice questionnaire was designed to gather information from 60 practicing veterinarians via Drag'n Survey. Out of forty questions, six were selected to assess the understanding, management, treatment efficacy, and prevention of Newcastle disease in the poultry industry. The results from this questionnaire provided valuable insights and met the study's objectives. Observations from veterinary practitioners indicated a 65.84% prevalence of Newcastle disease in broilers, with a subsequent rate of over 15% in laying hens, 11.53% in future laying hens, and 7.69% in broiler breeders. Our findings revealed an overall prevalence of 38.91% for Newcastle disease, with infectious bronchitis following at an average prevalence rate of 32%. Gumboro disease had a prevalence of 20%, and weak avian influenza was reported with a frequency of 12%. Infectious laryngotracheitis was the least common, with an average prevalence rate of 4%.

Keywords: Newcastle disease; statistical model, chai chicken; clinical manifestations; lesions; inhibition of hemagglutination; Algeria.

I. INTRODUCTION

The broiler poultry sector is the largest and most efficient meat production industry in the world (Gupta et al., 2014). In Algeria, as in many other countries, broiler production is threatened by a variety of infectious diseases, particularly viral ones. These diseases result in significant economic losses, with no reliable medication currently available (Pradhan et al., 2014). Newcastle Disease (ND) is the most economically significant poultry disease, especially in developing countries, due to its high mortality rates and the extensive sanitary measures required in poultry farms and slaughterhouses (Ban-Bo et al., 2013). It is essential to study the epidemiological characteristics of Newcastle Disease and to develop a program to combat this pathology. Samples collected from poultry farms are used for both animal health control (e.g., samples from poultry, autopsies, tracheal swabs, blood samples) and public health monitoring. ND is caused by virulent strains of avian paramyxovirus type 1 (APMV1) and is characterized by significant variability in morbidity, mortality, clinical signs, and lesions. This virus is highly contagious among all age groups and can infect numerous species of domestic and wild birds (Brown et al., 1999). Biosecurity and agricultural practices play a crucial role in the severity of the disease observed on affected farms (Jaganathan et al., 2015).

The hemagglutination inhibition (HI) test is a diagnostic tool for Newcastle Disease and provides information about vaccine immunity (De Langhe &Jorna, 2006). It is a reference technique in serology. Hemagglutination inhibition is used in immunology to detect specific antibodies in a biological sample. When specific antibodies are present, they prevent agglutination (clumping of red blood cells) caused by an agglutinating agent, such as a virus or bacteria (Al-Garib et al., 2003). The outcome of the hemagglutination inhibition test depends on whether agglutination occurs. If agglutination occurs, it indicates that specific antibodies are absent and the agglutinating agent has acted freely on the red blood cells. Conversely, if no agglutination occurs, it indicates the presence of specific antibodies in the sample, which have inhibited agglutination by binding to the agglutinating agent.

In summary, a positive hemagglutination inhibition result signifies the presence of specific antibodies, while a negative result indicates their absence (Al-Garib et al., 2003).

In statistical analysis, a range of tests and methods are used to examine the relationships between variables. In this study, the chi-square test of independence was employed to assess the association between categorical variables (McHugh, 2013). Pearson's correlation coefficient was used to measure the linear relationship between continuous variables (Schober et al., 2018), while Cramer's V was utilized to evaluate the strength of association between nominal variables (Akoglu, 2018). Additionally, multiple regression analysis was applied to examine the relationship between a continuous dependent variable and multiple independent variables (Hidalgo & Goodman, 2013). The p-value was used to determine the statistical significance of the results, considering the context and limitations of p-values in scientific research (Wasserstein & Lazar, 2016). By applying these statistical methods, this study aims to provide valuable insights into the factors associated with disease prevalence in poultry farms and identify potential interventions for managing these diseases.

II. MATERIALS AND METHODS

II.1 Sites and period of experimentation

Our selection of sites in various regions of Algeria was based on several key factors, including the cooperation of veterinary practitioners, the efficiency of the circuit, the high number of animals per site (exceeding 20,000 per building), and the accessibility of the sites throughout all seasons. The study was conducted from April 2023 to April 2024. We collected samples from thirty private broiler poultry farms that were clinically affected by a viral disease and exhibited characteristic lesions upon necropsy examination. Forty industrial-type broiler farms, each housing between 20,000 and 70,000 animals and aged 4 to 7 weeks, were selected (with 15 animals sampled per farm). Veterinarians responsible for monitoring reported suspected cases and collected two sets of samples from each farm. The first sample was collected early, within one to two days from the onset of infection, and the second sample was collected later, two to three weeks thereafter.

Alongside sampling, zootechnical and health data were collected through interviews with farmers, the monitoring veterinarians (regarding lesions and suspicions), or direct observation. A data sheet identifying each farm and a monitoring sheet documenting the evolution of the livestock's general condition were prepared. For the poultry health situation, the following information was sought:

- The occurrence and frequency of Newcastle disease throughout the year;
- The livestock and age groups most affected;
- The description of clinical and lesional signs;
- The climatic conditions and the suspected season or period of Newcastle disease outbreaks;
- The morbidity rate and associated mortality;
- The vaccination protocol (including the age of vaccination, type and method of vaccine administration, relapse cases, and vaccine quality as indicated by viral passage).

After autopsies on fresh or sacrificed cadavers, organs (trachea and cloaca) were removed and sent to the laboratory, where they were centrifuged the same day (5000 rpm for 10 minutes) to collect sera. These sera were then stored in Eppendorf tubes, identified, and frozen at -20 °C. The samples were sent to the nearest Approved Analysis Laboratory (LDA), contingent upon prior verification of its operational status. Official approvals for virological or serological diagnosis of Newcastle disease are issued by the Ministry of Agriculture and communicated to the Departmental Directorates of Veterinary Services (DDSV), where they are subsequently confirmed and completed by the National Reference Laboratory (NRL).

II.2 Laboratory diagnostic techniques

The confirmation of Newcastle disease was primarily achieved through the isolation of the virus from 9- to 11-day-old embryonated eggs. A sample is considered positive for the presence of the virus if it demonstrates a positive result in the rapid hemagglutination test. The virus is identified by its inhibition of hemagglutination (Ichakou, 2004). The hemagglutination inhibition test was conducted using 96-well plates made of high-quality optical polystyrene (Greiner Bio-One). In this test, the hemagglutination (HA) antigen is mixed with serially diluted antisera and incubated. Following this, a fixed concentration of red blood cells is added to each well, and the degree of antibody binding to the HA molecules is assessed. Data from the survey and laboratory analyses were analyzed using Excel and SPSS software.

III. RESULTS AND DISCUSSION

III. 1. IN THE FIELD DIAGNOSTIC

III.1.1 Livestock most affected of the samples taken

Observations of Newcastle disease reported by practicing veterinarians indicate a diagnosis frequency of 65.84% in broilers. This is followed by a rate exceeding 15% in laying hens, a prevalence of 11.53% in future laying hens, and a prevalence of 7.69% in broiler reproduction (see Table 1).

Table 1.Most affected farm of the samples collected in percentage.

Broilers	Laying hens	Future laying hens	Broiler Production
65.84%	15.00 %	11.53%	07.69%

Table 1 presents data on the prevalence of affected subjects. It is observed that broiler chickens exhibit a higher susceptibility to pathology, with an incidence rate of 65.84%. Among the age groups, the growth phase is the most affected, accounting for 46.87% of the cases. This is followed by the finishing phase, which represents 43.75% of the cases, and the start-up phase, which shows a prevalence rate of 9.37% (see Figure 1).

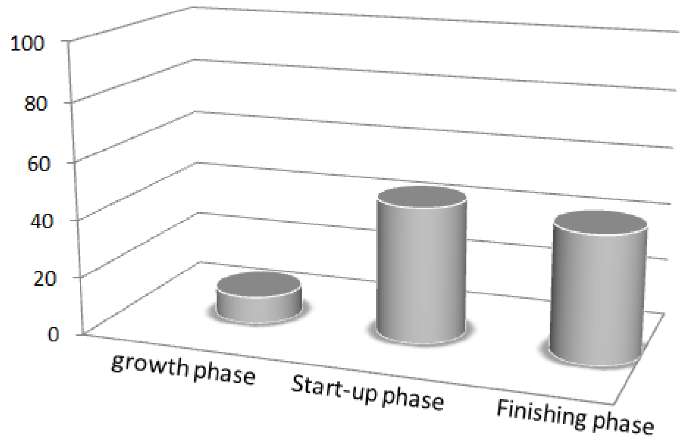


Figure 1. Most affected age group in the farms visited.

Our results indicate that during the growth phase, the disease affects 46.87% of the livestock. This prevalence is lower compared to the findings of Kermiche and Kaouche (2019), who reported that the disease reaches its peak during the growth phase, affecting 75% of the livestock. These observations are consistent with the documented prevalence of Newcastle disease. Broilers are particularly vulnerable to this pathology, with a prevalence of 65.84% during the start-up phase (from 1 to 15 days). The prevalence rate during this phase is 9.37%, followed by 48.87% in the growth phase, and 43.75% in the finishing phase (from 30 to 60 days), as illustrated in Figure 1.

III.1.2 The most common diseases in broiler farming

Out of a total of 116 diseases, the highest mortality rate is recorded in viral diseases with 31.03%, 24.13% mortality rate caused by bacterial diseases, 12.41% by parasitic diseases and a rate of 2.41% caused by diet-related diseases (Figure 2).

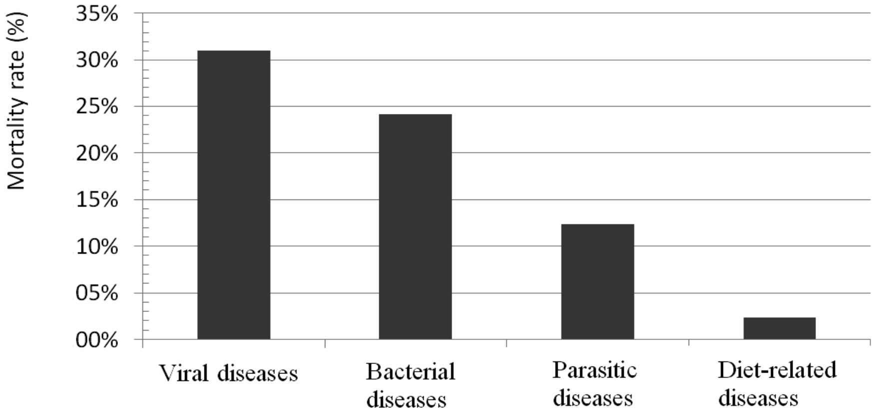


Figure 2. Diseases causing the highest mortality rate.

Our results on viral diseases (31.03%) are much lower than those found by (Rezki, 2014), where viral diseases are at 44.52% and Kermiche and Kaouche (2019) with a prevalence of 65%. In Figure 2, we have noticed that mortality is related to the type of disease that attacks the livestock. A mortality rate is around 31.03%, for viral diseases, followed by a frequency of 24.13% caused by bacterial diseases, 12.41% caused by parasitic diseases and. On the other hand, a mortality of 2.41% recorded by diet-related diseases.

III.1.3 Viral diseases according to suspicion

The average prevalence of infectious bronchitis is approximately 32%, while Newcastle disease has a prevalence of 38.91%, with some reports indicating up to 100%. Gumboro disease ranks third with a prevalence of 20%, followed by weak avian influenza at 12%. Infectious laryngotracheitis has the lowest prevalence among the listed diseases, with an average rate of 4% (see Figure 3).

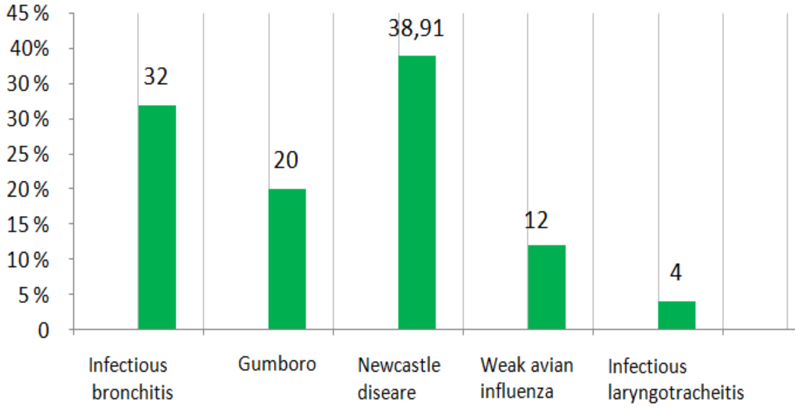


Figure 3. Viral diseases by suspicion

III.2. Description of clinical and lesional signs

III.2.1 Symptoms observed in an affected flock

The symptoms found on 112 affected farms are distributed as follows: Prostrations and depression with a percentage of 28.57, Ruffled feathers with a percentage of 23.21, Torticollis with a percentage of 32.14, Paralysis (legs, wings or other parts of the body) with 16.07% (Figure 4).

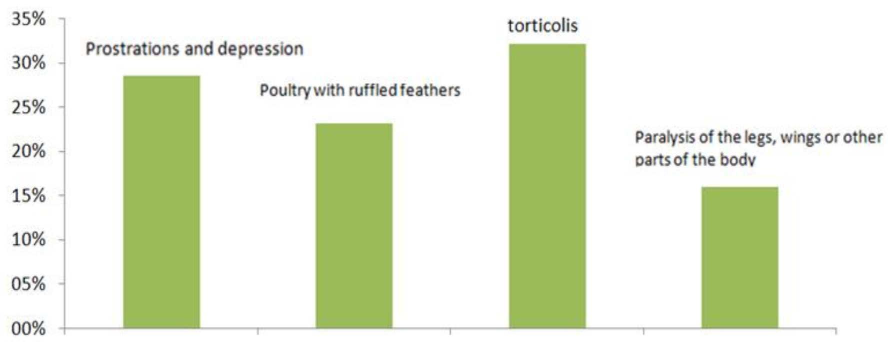


Figure 4: Symptoms observed in an affected flock.

III.2.2 Clinical and lesional manifestations in an affected flock

At necropsy, clinical and lesional manifestations included respiratory signs in 28% of cases (panting, coughing, sneezing, and rales), with corresponding respiratory lesions observed in 26%. These lesions were characterized by swelling of the head or periorbital region and edema in the interstitial tissue of the neck. Our results (28%) are lower than those reported by Rezki (2014), who found a prevalence of respiratory manifestations at 44.14%. Nervous signs, including tremors, circling, clonic spasms, wing and leg paralysis, and torticollis, were also noted, with nerve damage observed in 14% of cases (see Figure 5). Renal tropism was indicated in 4% of cases, with kidney lesions present in 6%. Digestive signs were present in 24% of cases, and digestive lesions were observed in 34%, consistent with Rezki's (2014) findings of 34.23%. Additionally, other clinical signs included partial or complete cessation of egg production, with eggs showing possible abnormalities in color, shape, or surface, and containing liquid albumin. Mortality rates werehighly variable but couldreach 100%.

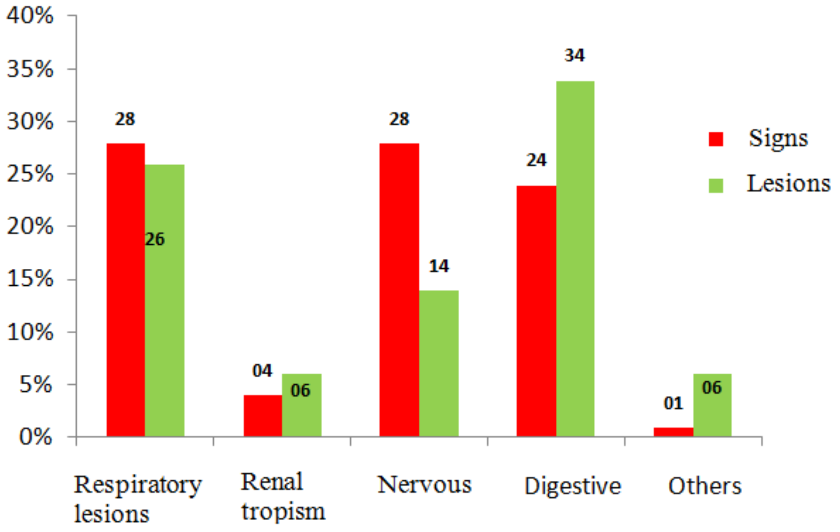


Figure 5. Clinical and lesional manifestations in an affected flock

Digestive signs with a percentage of 28.57% with digestive lesions with a percentage of 39.53% of which 50% are represented by petechiae in the proventricle and 24% hemorrhages in the intestine 26% mucus in the trachea (tracheitis) (Figure 6).

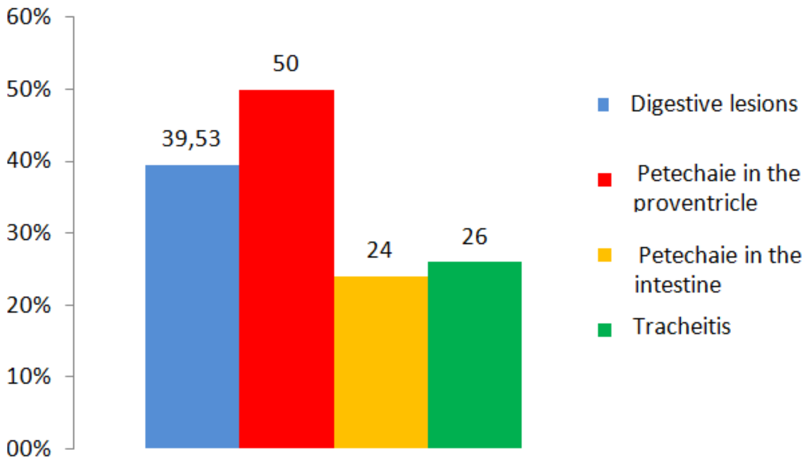


Figure 6. Different digestive lesions encountered.

Other symptoms were also observed, with the most prevalent being torticollis (90%), prostrations, and depression (85%). Additionally, 65% of poultry exhibited ruffled feathers, and 45% experienced paralysis of the legs, wings, or other body parts. Other clinical signs noted by practicing veterinarians include loss of appetite (98%), despondency (88%), runny nose (80%), decreased egg-laying (100%), and mortality (100%) (Figure 7). Our findings align with those reported by Ebwa (2019), who documented a 100% frequency of hen mortality and a decrease in egg-laying, with loss of appetite at 99%, fever at 98%, plumage dragging on the ground at 97%, bloody diarrhea at 94%, head raising at 90%, despondency at 88%, greenish diarrhea at 82%, runny nose at 80%, crust on the head at 79%, mortality of palmipeds at 77%, white diarrhea at 74%, yellow diarrhea at 65%, convulsions at 13%, and cyanosis at 8%. Both acute and chronic clinical forms of the disease were evident in these results. Initially, hens exhibited fever along with whitish or greenish diarrhea and occasionally lost their appetite. Subsequently, they would become debilitated, with their plumage dragging on the ground, and eventually succumb to the disease (Ebwa, 2019).

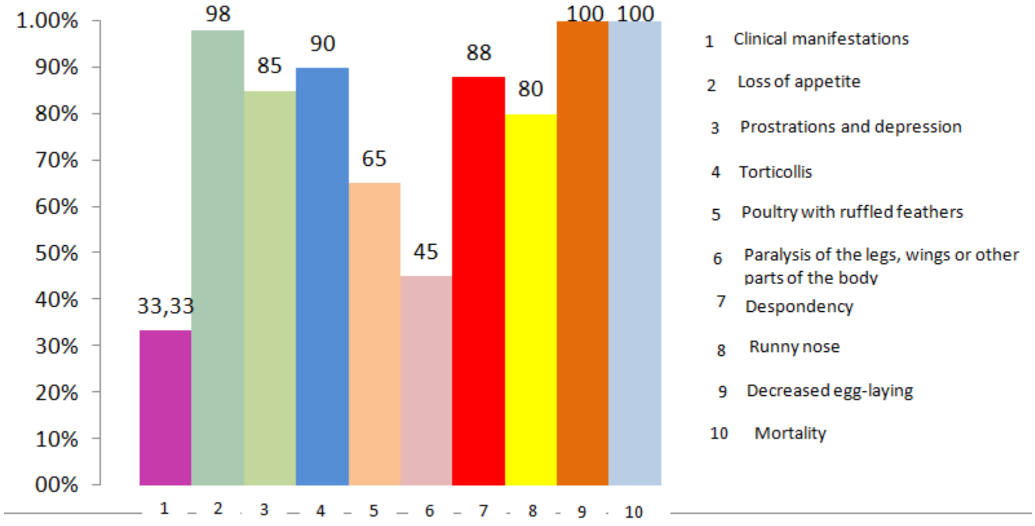


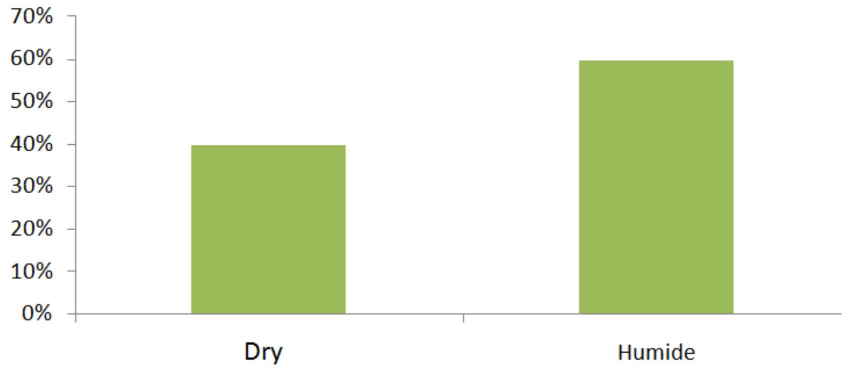
Figure 7. Clinical manifestations in affected individuals

According to Rauw et al. (2009), the incubation period of Newcastle disease virus ranges from 2 to 15 days. Common clinical signs of Newcastle disease (ND) include depression, diarrhea, prostration, edema around the eyes, and a reduction or cessation in egg production. Mortality and neurological symptoms, such as torticollis and clonic convulsions, are frequently observed. Hemorrhagic lesions in the proventriculus and intestines are also commonly present.

III.2.3 The distribution of livestock farms according to climate

Our observations indicate that Newcastle disease prevalence is significantly higher during wet periods, occurring in 60% of cases, compared to a 40% reduction during dry periods (Figure 8). The elevated temperatures characteristic of the hot and dry season can substantially inactivate viruses and thereby reduce the incidence of Newcastle disease. Conversely, the cold and dry season appears to create optimal conditions for the pathogen's spatial distribution, as noted by Ban-Bo et al. (2013). Factors such as cooling, draughts, and exposure to rainfall in open fields or inadequately protected poultry houses act as stressors that facilitate the outbreak of Newcastle disease (Jaganathan, 2015). The seasonal variation has a notable impact on the progression of the disease, which frequently assumes an epizootic character during the dry and windy season (Rezki et al., 2014). According to Ban-Bo et al. (2013), the dry season, extending from October to April, is particularly influential. This period is characterized by a transition from dry-cool winds in October-November to dry-hot, dusty winds in March-April, a climatic shift that appears to play a critical role in the manifestation of Newcastle disease.

Figure 8. Distribution of livestock by climate.



III.2.4 Theseasonal distribution of Newcastle disease

The annual epidemiological calendar and the results from the questionnaires indicate that, among 92 cases, there was an epizootic peak during the summer season, characterized by a high incidence with an average temperature of 29°C. This season exhibited a prevalence rate of 32.96% (30/92 cases). In contrast, other seasons recorded lower prevalence rates: winter and spring, with an average temperature of 7°C, had a prevalence rate of 23.91%, while the fall season, with an average temperature of 16°C, had a prevalence rate of 19.56% (see Figure 9).

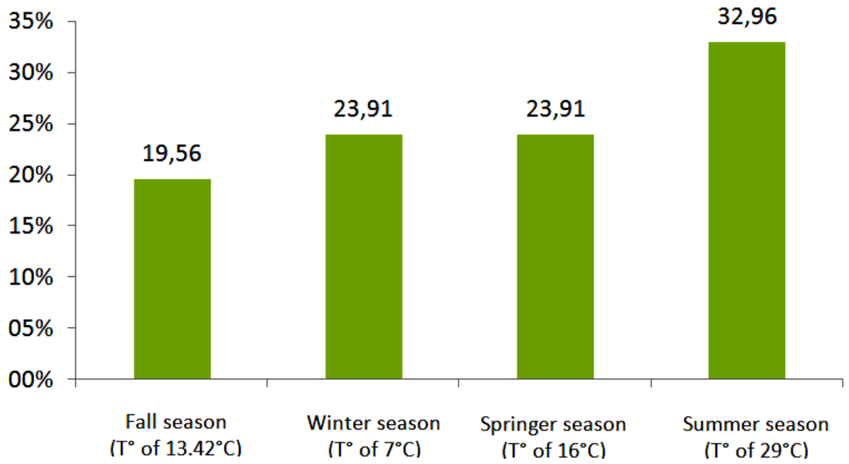


Figure 9. Seasonal distribution of Newcastle disease

Ebwa (2020) observed that during the summer period (June to July), the incidence of the disease peaks in June (85%) and July (73%), a time characterized by very high animal mortality. Similarly, Maminiaina (2007) documented a disease in Newcastle, Madagascar, exhibiting both enzootic and epizootic patterns, with a peak in October. Additionally, Rasamoelina (2011) reported the same disease in Lake Alaotra, Madagascar, with an epizootic peak occurring between August and October.

III.3 The vaccination protocol

III.3.1 Vaccination

Out of 80 reasons that can cause this pathology, it has been observed : A vaccine failure with a percentage of 34%. An unsuitable vaccine strain with a percentage of 12%**.** An unsuitable vaccination program with a percentage of 26% . Other undefined reasons with a percentage of 8%.

Table 2: Vaccination protocol and other accompanied manifestations.

Protocol	Types	Total cases	Overall percentage (%)	Observations
Vaccination	Failure vaccination Unsuitable vaccine strain Unsuitable vaccination program Others	80	34 12 26 8	
Existence of a vaccination protocol	National Personnel Use the laboratory	22 28 4	20 70 10	
State of hygiene	Good Medium Poor	30	23.3 30 4.6	
Density	n< 3000 3000 <n< 4000 n>4000	29 farms	13.5 36.6 50.00	The stocking density is 10 chickens/m². [Louer, Bellaoui]
Mobility rate	0-50 50-100		50 50	morbidity 8-10 % Mortality 5.7-82% and case fatality rates 10-100% [SaliouSylla et *al.*, (2003)]
Manifestations accompanied by mortality	n< 30 30<n< 70 70<n< 100	40 farms	60 35 5	

Despite the availability of vaccines, Newcastle disease (ND) remains a significant challenge in poultry farming in West Africa (Eze, 2014). Vaccine failure occurs in approximately 34% of cases, which may be attributed to the high resistance of local poultry. Additionally, the limited mastery of diagnostic methods complicates the accurate identification of the disease, often leading to its misdiagnosis as other viral infections (Ichakou, 2004). This misdiagnosis hinders effective decision-making. Furthermore, many farmers lack knowledge of alternative control methods beyond vaccination, which is a major limiting factor in managing Newcastle disease. Inappropriate vaccination programs account for 26% of the failure rate. For unsuitable vaccine strains, a 12% failure rate has been recorded, and the remaining 8% are attributed to unknown causes. Incorrect vaccine application is identified as one of the most common reasons for vaccination campaign failures (Marangon et al., 2006). The success of vaccination is significantly influenced by the level of maternally derived antibodies (MDAs) in young poultry, which can vary widely among farms, chick batches, and individuals. Other undefined factors contributing to the 8% of Newcastle disease cases (see Table 2) may include farm hygiene and stocking practices. According to Miller et al. (2007), a vaccine strain that is phylogenetically distant from circulating strains does not offer optimal protection, in terms of wild virus shedding, compared to a homologous vaccine strain.

III.3.2 Existence of a vaccination protocol

Out of 40 surveyed veterinarians, all reported using a vaccination protocol. Among 54 responses regarding the types of protocols employed, three distinct categories were identified: National Protocol, Personal Protocol, and Use of the Laboratory. Specifically, 20% of respondents (22/54) adhered to the National Protocol, 70% (28/54) followed the Personal Protocol, and 10% (4/54) utilized the laboratory. The predominance of the Personal Protocol, which accounts for 70% of responses, indicates a situation where consultation with the veterinarian or laboratory use is not commonly practiced. This observation is corroborated by Allan and Dawson (1973), who noted a high mortality rate associated with infrequent vaccination.

III.3.3 Cases of relapse after vaccination

According to the veterinarians interviewed, 50% of vaccinated poultry farms experience disease outbreaks. This can be attributed to the use of attenuated viral strains in the most commonly used commercial vaccines. Contributing factors include vaccine failure, which may result from improper storage or expired vaccines, and a lack of adherence to vaccination protocols. Kermiche and Kaouche (2019) report a slightly higher incidence of 65.73%. Conversely, 50% of veterinarians reported no disease occurrences, which is likely due to the absence of vaccination, highlighting its role as a preventive measure against disease. Research by Viaene et al. (1972) and earlier studies identified a velogenic strain of Newcastle disease isolated in Belgium in 1971 (isolate Gent, 7I). The antigenic properties of this strain were found to be very similar, if not identical, to those of commonly used vaccine strains. Maternal antibodies (HI 40) were insufficient to protect chicks from experimental infection (Viaene et al., 1972b). However, vaccinations administered to very young chicks with similar levels of maternal antibodies (HI 40) successfully protected these chicks from severe experimental infections. These findings underscore the importance of early vaccination in chicks and help explain occurrences of avian pseudopest after vaccination, particularly when chicks are exposed to virulent infections shortly after arrival on the farm and during their early days of life.

III.3.4 Hygiene

According to the questionnaire responses, 7 out of 30 farms (23.3%) were classified in the Good Hygiene category, 9 out of 30 farms (30%) were categorized as Intermediate Hygiene, and 14 out of 30 farms (46.6%) were placed in the Poor Hygiene category. Biosecurity, encompassing a comprehensive set of stringent sanitary and hygienic measures, represents the most effective and cost-efficient approach for controlling poultry health over the long term. It is crucial for enhancing farm profitability and product quality. As the primary defense against the introduction of avian diseases, particularly Newcastle Disease (ND), biosecurity measures play a critical role in safeguarding poultry health (Bermudez, 2003; Bermudez et al., 2003).

III.3.5 Density

In a study conducted on 29 farms categorized based on animal density per building ($n < 3000$, $3000 \leq n \leq 4000$, and $n > 4000$), the distribution of farms was 13.3% (4/29), 36.6% (11/29), and 50% (15/29), respectively. According to Laouer (1987) and Bellaoui (1990), the recommended stocking density is 10 chickens/m². The incidence of disease reaches 50% in populations exceeding 4000 individuals, regardless of age. Alexander (2003) reports that poultry surviving an epizootic disease can become carriers of Newcastle disease. Additionally, the virus has been shown to persist in poultry and adapt to environmental conditions (Alexander, 2001). While young chickens typically lose their maternal antibodies within one month, rendering them susceptible to infection, surviving hens may continue to shed the virus (Ban-Bo et al., 2013). This persistence contributes to the prevalence of Newcastle disease within the epidemiological unit. Following an epizootic, survivors often exhibit resistance to subsequent Newcastle disease infections. Producers, recognizing this trait, frequently obtain these resilient birds from weekly markets or through donations to replenish their flocks. The primary method for restocking poultry yards is through reproduction (Koko et al., 2002).

III.3.6 Morbidity and mortality rates

Morbidity and mortality rates vary considerably depending on factors such as the animal's immunity, the strain's virulence, host susceptibility, and other environmental conditions. For velogenic isolates, morbidity and mortality rates can reach up to 100% in fully susceptible, unvaccinated chickens. In contrast, in healthy poultry, the mortality rate is approximately 10% for mesogenic strains and negligible for lentogenic strains. Estimates from 50% of practicing veterinarians suggest that the morbidity rate in Newcastle disease varies significantly, ranging from 0% to 50% and from 50% to 100%. The economic impact of this viral disease is substantial due to its high mortality and morbidity rates and its consequences on international trade, posing a severe burden on livestock farms (Ban-Bo et al., 2013). According to SaliouSylla et al. (2003), morbidity, mortality, and case fatality rates ranged from 8% to 100%, 5.7% to 82%, and 10% to 100%, respectively.

III.3.7 Manifestations accompanied by mortality

Out of 40 responses, it was unanimously observed that all clinical manifestations of Newcastle disease are accompanied by mortality. The farms were categorized into three groups based on the mortality rate associated with the disease: 0% to 30%, 30% to 70%, and 70% to 100%. The survey results indicated that 60% of the farms (24 out of 40) experienced a mortality rate within the 0% to 30% range, 35% of the farms (14 out of 40) reported a mortality rate between 30% and 70%, and 5% of the farms (2 out of 40) had a mortality rate ranging from 70% to 100%. According to the veterinarians surveyed, 60% reported that the mortality rate on farms affected by Newcastle disease ranged from 0% to 30%, 35% indicated a range of 30% to 70%, and 5% reported a range of 70% to 100%.

III.4. STATISTICAL ANALYSIS

A comprehensive statistical analysis was conducted to explore the relationships between various factors influencing disease prevalence in poultry farms, utilizing data from multiple sources. These factors included livestock type, age group, disease type, symptoms, clinical and lesional manifestations, and environmental conditions such as climate. A range of statistical tests was employed to assess these relationships, including the chi-square test of independence, Pearson's correlation coefficient, and Cramer's V test.

Key Findings:

1. **Livestock Type and Disease Prevalence:** The chi-square test revealed a strong association between livestock type and disease prevalence ($V = 0.6$), indicating that susceptibility to the disease varies among different types of livestock.

2. **Age Group and Disease Prevalence:** A moderate association was found between age group and disease prevalence ($V = 0.4$), suggesting that different age groups exhibit varying levels of vulnerability to the disease.

3. **Disease Type and Prevalence:** There was a strong association between disease type and prevalence ($V = 0.5$), implying that different disease types have varying occurrence rates and impacts.

4. **Specific Viral Diseases and Suspected Prevalence:** A very strong association was observed between specific viral diseases and their suspected prevalence ($V = 0.7$), indicating that the suspicion of these diseases is a reliable indicator of their presence.

5. **Symptoms and Disease Prevalence:** Multiple regression analysis revealed that symptoms such as prostration and depression, ruffled feathers, torticollis, and paralysis significantly predict disease prevalence (adjusted $R^2 = 0.6$), suggesting that these symptoms are useful in identifying and managing the disease.

6. **Clinical and Lesional Manifestations:** Clinical and lesional manifestations (including respiratory, nervous, renal, and digestive signs) were significant predictors of disease prevalence (adjusted $R^2 = 0.7$), highlighting their importance as diagnostic markers.

7. **Climate and Disease Prevalence:** A weak to moderate association was found between climate and disease prevalence ($V = 0.3$), implying that while climate may influence disease occurrence, other factors might play a more significant role.

8. **Temperature and Newcastle Disease Incidence:** Pearson's correlation analysis showed a moderate positive correlation between temperature and Newcastle disease incidence ($r = 0.702$), but this relationship was not statistically significant ($p = 0.298$).

9. **Hygiene, Animal Density, and Morbidity/Mortality Rates:** A strong positive relationship was observed between hygiene and both animal density ($r = 0.940$) and morbidity/mortality rates ($r = 0.751$), as well as a moderate positive relationship with clinical manifestations and mortality ($r = 0.632$). Pearson's correlation tests in R confirmed the statistical significance of these relationships at a 0.05 significance level.

The statistical analysis demonstrated several important associations between various factors related to disease prevalence in poultry farms, including livestock type, age group, disease type, symptoms, clinical and lesional manifestations, and hygiene. These findings suggest that controlling these factors, such as improving hygiene conditions and reducing high animal density, can help mitigate disease spread and its negative impacts. However, further studies should be conducted to confirm these associations and identify the most effective interventions for managing diseases in specific contexts.

CONCLUSION

At the conclusion of this study on the diagnosis of Newcastle disease, several critical insights were highlighted. The data revealed that 65.84% of broiler farms were affected by Newcastle disease, with an overall prevalence rate of 38.91%. The clinical manifestations of the disease are influenced by a range of factors, including the specific virus strain, host characteristics, age of the host, concurrent infections by other microorganisms, environmental stresses, and immune status. While clinical signs combined with autopsy can provide valuable diagnostic information, laboratory confirmation remains essential for accurate diagnosis.

Our findings underscore the significant role of biosecurity measures and agricultural practices in influencing the severity of Newcastle disease. Effective farm management involves prompt response to suspected cases of Newcastle disease and adherence to the measures outlined in community directives, which significantly reduce the risk of viral transmission.

Despite vaccination efforts, the questionnaire results indicated that suspicious cases of Newcastle disease are often confirmed positive upon further testing, suggesting the possible emergence of a new viral variant. This situation necessitates the development of new strategies and formulations to combat the disease, which continues to pose a challenge to poultry farming. Although vaccination is crucial and mandatory, farmer reluctance due to the associated workload remains a concern. The use of inactivated vaccines could enhance the defense of susceptible poultry.

One notable characteristic of the APMV-1 virus is the considerable variation in pathogenicity among different viral strains. Contaminated poultry products are recognized as risk factors for the transmission of Newcastle disease.

RECOMMENDATIONS

Serology alone does not explain the level of protection induced by vaccination. As previously stated, currently marketed vaccines may lack efficacy and have some drawbacks of use. The implementation of molecular biology techniques in the field of vaccination aims to improve the effectiveness and safety of conventional vaccines. It is therefore necessary to develop an "ideal vaccine" capable of protecting animals from the disease and inhibiting the spread of the virus during infection, while limiting the workload for farmers. These new techniques will provide a better understanding of the mechanisms of cell-mediated immunity and the local immune response (in the respiratory and digestive tract) specific to Newcastle disease and their role in protecting against clinical signs and virus shedding. Given the importance of Newcastle disease, there is an urgent need to offer endogenous alternatives through the development of plants that are usually little used in modern chicken farming. These are low-cost and highly available resources.

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